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STUDIES IN THE CHEMISTRY AND PHYSIOLOGY OF THE LEAVES OF THE BETEL-VINE (*PIPER BETLE*) AND OF THE COMMERCIAL BLEACHING OF BETEL-VINE LEAVES

PART II

BY

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INTRODUCTION.

In July 1913 we published in the present series of Memoirs[1] an account of investigations which we had made into certain questions concerned with the growth and utilization of the betel-vine leaf which, as is well known, forms probably the most important garden crop of India and is also one of the most intensive cultures of the country. We then discussed three questions, namely, (1) the occurrence of nitrates in betel-vine leaves and their relationship to the growth of the vine, (2) the sugars, starch, tannin, essential oil, and other normal constituents of the betel-vine leaf, and their relationship to the growth of the vine, and (3) the commercial bleaching of the betel-vine leaf, and the chemical changes by which it is accompanied. We have since carried our investigations on these questions considerably further and have moreover had the opportunity to examine in more detail than has hitherto been done, the nature of the essential oil, which, from the point of view of the commercial value of the leaf, appears to be the most important constituent of the product. The following is an account of the principal results obtained in these directions.

[1] *Memoirs of the Dept. Agric. India, Chem. Series*, vol. III, no. 2 (1913).

I.

The Normal Constituents of the Betel-Vine Leaf and their Relationship to the Growth of the Vine.

The question which led to our study of the variation of the normal constituents of betel-vine leaves at different periods of their growth, and at various times in the day was really that of ascertaining the cause of the use of the leaf for chewing purposes. As we noted in our former memoir, no appreciable quantity of an alkaloid and no active glucoside seem to be present in the leaf. To what extent therefore the quantity of the other constituents differs at various times in the life of the plant, and what relationship each of them bears to the value of the leaf was the problem before us. In continuation of our former work the following normal constituents have been investigated :— (1) the tannin, (2) the sugars, (3) the starch, (4) the ether extract, (5) the essential oil, (6) the acidity, (7) the diastatic activity, (8) the nitrates. The results obtained and previously published[1] indicated that the leaves near the top of the plants contain much more essential oil,[2] much more diastase, and much more sugars than those which are lower on the vine. The tannin did not vary in this direction ; the leaves of full age in the middle of the vine, neither in the young and actively growing part nor in the older leaves near the base of the plants, contained the largest quantity of this constituent. Further, the constituent which seemed to vary most closely with the market value of the leaves was the essential oil. The present paper carries these results considerably further.

The methods of estimation of these constituents were those employed and described in our previous paper.[3]

We will first consider the variation of the constituents at different parts of the day in the leaves taken from a normal vine, from which the leaves were being plucked for sale. The samples were taken a few days indeed before the annual operation of taking down the vine, coiling it up and burying it, and allowing it to send out the new shoots, which gradually grow and yield the vines of the succeeding year. This time represents the leaves slightly later than at their best from a commercial point of view.

[1] *Loc. cit.*

[2] It was erroneously concluded in our previous memoir that the results obtained were connected with the age of the leaves ; our later results indicate clearly that it is the position on the plant and hence possibly the relation to light that determines the proportions of these constituents.

[3] *Loc. cit.*

Under these circumstances leaves were taken from adjacent vines in the same garden. The leaves were quite mature, fully developed, rather dark in colour, slightly brittle, and with their tips slightly dried. They had been on the vine a little longer than is wise to give absolutely the best results on bleaching, but still they were quite sound and satisfactory for this purpose. Samples were taken on three successive days, taking three samples every day—the first in the morning between 7 and 8 A.M., the second in the afternoon between 1 and 1·30 P.M., and the third in the evening between 6 and 7 P.M.

The results obtained in this series, in view of the fact that leaves for sale are generally plucked chiefly in the morning, are very interesting, and are shown in the following table. All figures are calculated as percentages of the dry matter of the leaves, except the acidity, and the diastatic activity.

	Reducing sugars	Non reducing sugars	Total sugars	Starch	Tannin	Ether extract	Essential oil	Nitrates (as KNO_3)	* Acidity	† Diastatic activity
	%	%	%	%	%	%	%	%	cc.	grm.
First day.										
Morning leaves ...	0·68	2·65	3·33	0·62	0·91	13·4	1·23	1·77	8·50	4·70
Midday leaves ...	0·90	2·39	3·29	0·87	1·18	12·1	1·35	1·52	8·50	10·20
Evening leaves ...	1·35	2·55	3·91	1·20	1·18	12·7	1·09	1·36	9·00	5·66
Second day.										
Morning leaves ...	0·71	2·72	3·43	0·70	1·11			1·89	8·50	4·22
Midday leaves ...	0·81	2·50	3·31	0·67	1·20			1·79	9·00	9·48
Evening leaves ...	1·20	2·67	3·87	1·10	1·33			1·65	9·50	7·60
Third day.										
Morning leaves ...	0·76	2·57	3·33	0·80	1·09			1·77	8·00	6·54
Midday leaves ...	0·62	2·29	2·91	0·83	1·18			1·60	8·90	9·42
Evening leaves ...	1·27	2·18	3·45	1·24	1·31			1·43	9·40	6·24

* Cubic centimetres of N/10 Caustic Potash required to neutralise ten grammes of dry leaf.

† Maltose produced by ten grammes of dry leaves in 48 hours.

The average of the three days records gives figures as follows :—

	Reducing sugars	Non-reducing sugars	Total sugars	Starch	Tannin	Ether extract	Essential oil	Nitrates	Acidity	Diastatic activity
	%	%	%	%	%	%	%	%	cc.	grm.
Morning leaves ...	0·72	2·65	3·36	0·71	1·04	13·4*	1·23*	1·81	8·33	5·1
Midday leaves ...	0·78	2·39	3·17	0·79	1·10	12·1*	1·35*	1·61	8·80	9·7
Evening leaves ...	1·27	2·47	3·74	1·18	1·27	12·7*	1·09*	1·48	9·30	6·5

* One determination only.

So far, therefore, as leaves in good condition for eating are concerned, the following results with regard to those constituents would seem to hold.

The reducing sugars and the starch are their lowest in the morning while the non-reducing sugars (presumably chiefly cane sugar) are very constant throughout the day. The change during the day follows somewhat the lines which have been shown to hold with other plants.[1] These are interesting, however, as under the conditions of growth of these leaves the sunshine rarely if ever reaches them. The tannin increases in amount during the day, and hence the leaves in the evening are slightly more astringent than in the morning. The variation in the amount of essential oil seems to be somewhat irregular, but from these figures it rises at first, and then declines toward the evening. The nitrates consistently decline in amount during the light hours of the day, while the acidity consistently increases. The diastatic activity varies curiously. It increases towards the middle of the day in each case, and then declines again towards evening—in one case going below the amount found in the early morning.

[1] Brown and Morris [*Journal Chem. Society*, Vol. 63 (1893), page 669], give following figures for Tropæolum, the days being bright and sunny.

	1st day.		2nd day.	
	5 A.M.	5 P.M.	9 A.M.	5 P.M.
Starch ...	1·23 %	4·59 %	3·24 %	4·22 %
Non-reducing sugars ...	4·65 %	3·86 %	4·91 %	8·02 %
Reducing sugars ...	5·14 %	5·72 %	6·80 %	5·19 %

If the fact that the leaves are chiefly gathered in the morning is anything more than a matter of convenience, then one would consider that what was required for eating was a leaf with little starch, tannin, nitrates and acidity, but as much essential oil and cane sugar as possible. This is undoubtedly true as regards the essential oil ; with regard to the other constituents the matter must remain in doubt.

So far we have dealt with leaves from similar adjacent vines at different times of the day. We have now to turn to a long series of similar determinations at various stages in the growth of similar and adjacent vines. The vines, as we have already described, are all pulled down at a certain season of the year—generally two months before the commencement of the rainy season—and the stems coiled and buried. From these the new vines arise and form the yielding plants for the following year. The experiments were made with leaves derived from such new vines vigorously growing from the stage when they had reached five to six feet high. In taking them the lower three feet of the vine was always left untouched as the leaves from this part are rarely plucked for use.

Between the third and the fifth foot in height of the vine, there were usually six to eight branches, and on every branch there were at least four leaves. Of these the two nearest the main vine are always small in size, and generally more or less misshapen in appearance. They are never used for bleaching and so in all that follows we have kept these separately from the next two. These two others were, of course, younger leaves but all that were taken were sufficiently mature for use. In the last set a third lot of two leaves was fit for use and was taken. In the present series, however, we are in every case dealing with *still* growing leaves, while in those considered above they are slightly over-mature.

In all five sets of samples were taken from vines adjacent to one another in the same gardens, of the same age and conditions of growth. They were taken on dates as follows :—

1st set	...	July 28, 1913.
2nd ,,	...	July 30, 1913.
3rd ,,	...	August 14, 1913.
4th ,,	...	September 5, 1913.
5th ,,	...	September 25, 1913.

On one day (September 5th) leaves were taken from the main vine at the same level as these branches to compare with the above.

The following results were obtained, considering the constituents in order :—

1. Moisture.

	Morning 7 - 8 A.M.	Noon	Evening 5 - 6 P.M.
	%	%	%
A. *Leaves (two) nearest the main vine.*			
1st set	83·5	82·5	85·0
2nd „	84·0	83·0	80·5
3rd „	83·0	81·0	85·4
4th „	83·0	82·5	83·0
5th „	82·0	82·5	83·0
B. *Leaves (two) further from the vine.*			
1st set	85·0	84·5	86·5
2nd „	85·5	89·0	86·0
3rd „	85·5	86·0	85·5
4th „	85·0	86·0	86·0
5th „	86·0	87·3	87·0
C. *Leaves (two) farthest from the vine.*			
5th set	85·5	86·5	84·5
D. *Leaves from the main vine.*			
4th set	84·5	85·5	86·0

These figures were all obtained in the rainy season, and are wonderfully constant. On the whole, curiously, the amount of water in the leaves is smallest in the morning, and it also tends to become greater in the later sets of leaves. But the moisture is, nevertheless, very constant indeed.

2. Sugars.

	Morning 7 – 8 A.M.			Noon			Evening 5 – 6 P.M.		
	Reducing sugars	Non-reducing sugars	Total sugars	Reducing sugars	Non-reducing sugars	Total sugars	Reducing sugars	Non-reducing sugars	Total sugars
	%	%	%	%	%	%	%	%	%
A. Leaves (two) nearest the main vine.									
1st set ...	1·61	0·24	1·85	1·80	0·20	2·00	2·50	0·50	3·00
2nd ,, ...	1·47	0·53	2·00	1·76	0·74	2·50	1·92	1·00	3·92
3rd ,, ...	1·75	0·56	2·31	1·37	0·82	2·19	1·39	0·87	2·26
4th ,, ...	1·47	0·48	1·95	1·17	0·49	1·66	1·35	0·39	1·74
5th ,, ...	1·33	0·14	1·47	0·95	0·27	1·22	1·33	0·40	1·73
B. Leaves (two) further from the main vine.									
1st set ...	0·72	0·28	1·05	1·43	0·66	2·09	1·59	0·64	2·23
2nd ,, ...	1·35	0·40	1·75	1·28	0·72	2·00	1·07	?	?
3rd ,, ...	1·53	0·59	2·12	1·23	0·82	2·05	1·60	0·84	2·44
4th ,, ...	1·43	0·32	1·75	1·61	0·65	2·26	1·40	0·92	2·32
5th ,, ...	1·00	0·14	1·14	1·30	0·19	1·49	1·13	0·30	1·43
C. Leaves (two) furthest from the vine.									
5th set ...	0·83	0·17	1·20	1·33	0·33	1·66	0·80	0·11	0·91
D. Leaves from the main vine.									
4th set ...	1·37	0·48	1·85	1·23	0·42	1·65	1·22	0·92	2·14

The results obtained from leaves at different parts of the day are interesting to compare with those previously given. So far as sets A and B are concerned, which are the only ones used for eating as a rule, the reducing sugars increase from morning to night, the non-reducing sugars do likewise, but in much greater proportion and taking an average of the ten sets of determinations we have the following : –

	Reducing sugars	Non-reducing sugars	Total sugars
	%	%	%
Morning	1·37	0·37	1·74
Noon	1·39	0·55	1·94
Evening	1·53	0·65*	2·34*

*Average of nine determinations only.

The older leaves on the main vine—which we have previously found abnormal in other respects—and the younger leaves at the end of the branch do not seem to follow the same rule, but the results given are from one set of determinations only, and we should hardly care to place too much reliance upon them.

During the ripening of the leaves which has more importance for our present point of view—the amount of sugars, both reducing and non-reducing, seem to rise to a maximum and then decline. Taking the average of the morning, noon, and evening determinations we have figures as follows. We have added an average figure for the first two sets, as they were only separated by two days.

	Reducing sugars	Non reducing sugars	Total sugars
	%	%	%
A. *Leaves (two) nearest the vine.*			
1st set ...	1·80 } 1·76	0·31 } ·53	2·28 } 2·54
2nd „ ...	1·72	0·76	2·81
3rd „ ...	1·50	0·75	2·25
4th „ ...	1·33	0·45	1·78
5th „ ...	1·20	0·27	1·47
B. *Leaves (two) further from the vine.*			
1st set ...	1·25 } 1·24	0·53 } ·54	1·79 } 1·83
2nd „ ...	1·23	0·56	1·87
3rd „ ...	1·45	0·75	2·20
4th „ ...	1·48	0·63	2·11
5th „ ...	1·14	0·21	1·35

The maximum occurs in the older leaves before it takes place in those further along the branches.

3. **Starch.** The determinations of starch were as follows :—

	Morning	Noon	Evening
	%	%	%
A. *Leaves (two) nearest the main vine.*			
1st set ...	1·03 } 1·09	1·36 } 1·41	1·47 } 1·44
2nd „ ...	1·15	1·47	1·42
3rd „ ...	1·31	1·41	2·13
4th „ ...	1·90	1·59	2·00
5th „ ...	1·57	1·47	1·59
B. *Leaves (two) further from the main vine.*			
1st set ...	1·25 } 1·28	1·52 } 1·41	1·65 } 1·49
2nd „ ...	1·32	1·30	1·32
3rd „ ...	1·56	1·58	2·00
4th „ ...	1·59	2·04	3·17
5th „ ...	1·54	1·84	2·16
C. *Leaves (two) furthest from the main vine.*			
5th set ...	1·45	1·80	2·08
D. *Leaves on the main vine.*			
4th set ...	1·60	2·28	2·08

The results in this case seem very clear. The starch increases consistently toward evening, as would be expected. The increase is not nearly so great as has been found in many cases in plants growing in the sun during the greater part of the day. In such cases Brown and Morris[1] found with *Tropæolum* that the starch increased from 1·23 to 4·59 per cent., and from 3·22 to 4·22 per cent. on two different days for the day as a whole. On the average we have increase as follows : –

	Leaves (two) nearest the vine	Leaves (two) further from the vine
	%	%
Morning ...	1·49	1·45
Noon ...	1·46	1·66
Evening ...	1·72	2·06

The starch in these leaves too tends to increase as the leaves get older, but again this probably works up to a limit, and then tends to decline again. The figures though not entirely clear, seem to indicate this, and the small quantity of starch in the much more mature leaves referred to on pages 283 and 4 would seem to justify this contention. If so, taken with the fact that for eating purposes, starch-containing leaves seem objectionable, it would seem one of the factors which prevent young leaves being used for eating as a rule if others are obtainable.

4. Tannin. The figures for tannin are as follows :–

	Morning	Noon	Evening
	%	%	%
	A. Leaves (two) nearest the main vine.		
1st set ...	1·17	1·31	1·30
2nd ,, ...	0·91	1·04	1·01
3rd ,, ...	1·44	1·17	1·43
4th ,, ...	1·96	2·06	2·35
5th ,, ...	1·56	1·43	1·31
	B. Leaves (two) further from the main vine.		
1st set ...	1·67	1·43	1·44
2nd ,, ...	1·31	1·17	1·21
3rd ,, ...	1·62	1·56	1·70
4th ,, ...	1·82	1·69	2·05
5th ,, ...	1·95	1·57	1·44
	C. Leaves (two) furthest from the main vine.		
5th set ...	2·08	1·82	1·50
	D. Leaves on the main vine.		
4th set ...	2·72	1·96	2·21

[1] *Journal Chem. Society*, Vol. 63 (1893), page 669.

Thus the variations in the amount of tannin during the day, and during the growth seem very great, and it is not at all easy to draw general conclusions. On the whole it is not clear that there is any variation in these actively growing leaves which is directly attributable to the period of the day, either in the leaves nearest the main vine or in those further away. The average of the figures above are as follows :—

	Leaves (two) nearest the vine	Leaves (two) further from the vine
	%	%
Morning	1·41	1·67
Noon	1·40	1·48
Evening	1·48	1·57

In the youngest leaves there seems a slight reduction in the evening but that seems the only conclusion we can make.

There does not, moreover, seem to be any decided tendency to increase in the amount of tannin in leaves occupying similar positions in the vine as the young leaves get older ; if anything there are signs of a rise as the leaves mature followed by a decline.

	Leaves (two) nearest the vine		Leaves (two) further from the vine	
	%		%	
1st set	1·26	1.13	1·51	1·37
2nd	0·99		1·23	
3rd ,, ...	1·35		1·63	
4th ,, ...	2·12		1·86	
5th ,, ...	1·43		1·65	

These figures rather tend to confirm the previous conclusion that as the leaves get more suitable for eating the amount of tannin tends to decline a little. The difference is not, however, very great.

5. Ether extract and Essential oil. Although the total ether extract has really no special meaning, being a miscellaneous collection of materials

extracted from the leaf, we report the figures with the very important determination of the essential oil :—

	Morning 7—8 a.m.		Noon		Evening 5—6 p.m.	
	Ether extract	Essential oil	Ether extract	Essential oil	Ether extract	Essential oil
	%	%	%	%	%	%
	A. Leaves (two) nearest the main vine.					
1st set	6·2	0·7	4·6	0·6	5·8	0·7
2nd „	8·6	1·4	6·6	1·4	7·6	1·2
3rd „	7·3	2·0	7·2	1·4	7·5	1·9
4th „	7·3	1·9	7·9	2·2	8·5	1·5
5th „	8·4	3·7	8·5	2·5	8·8	2·9
	B. Leaves (two) further from the main vine.					
1st set	5·5	0·6	6·0	0·6	5·9	0·7
2nd „	6·4	1·5	6.5	1·5	6·4	1·8
3rd „	6·3	1·5	6·4	2·0	7·7	2·3
4th „	6·4	2·5	7·3	2·4	6·8	2·3
5th „	8·3	4·3	8·8	2·7	8·9	4·9
	C. Leaves (two) furthest from the main vine.					
5th set	6·0	1·4	5·4	1·6	5·8	2·0
	D. Leaves from the main vine.					
4th set	5·7	1·6	6·8	1·9	7·4	2·0

We shall not further consider the total ether extract, but the essential oil presents some interesting problems.

(a) *Variation of essential oil during the day.*

	Leaves nearest vine	Leaves further from vine	Leaves furthest from vine	Leaves on main vine
			(one set only)	(one set only)
	%	%	%	%
Morning	1·9	2·1	1·4	1·6
Noon	1·6	1·8	1·6	1·9
Evening	1 6	2·4	2·0	2·0

The variation appears irregular and not clearly a direct consequence of the time of day.

(b) *Variation of essential oil of similar leaves during development.*

	Leaves nearest vine	Leaves further from vine
	%	%
1st and 2nd sets (average)	1·0	1·1
3rd set	1·8	1·9
4th „	1·9	2·4
5th „	3·0	4·0

The amount of essential oil increases in both sets, to a very considerable extent. The increase is not a function of the maturity alone, or else those nearest the vine would tend to contain the most, but the position on the branch is effective to some extent. It must not be forgotten that the leaves nearest the vine are not preferred for eating and not used at all for bleaching.

(c) *Variation of essential oil due to position on vine.*

	Leaves nearest vine	Leaves further from vine	Leaves furthest from vine	Leaves on main vine
	%	%	%	%
4th set	1·9	2.4	..	1·8
5th	3·0	4·0	1·7	..

The youngest leaves, hence, at the same time appear to be poorer in essential oil than the others. The constituent quickly increases, however, and a maximum would appear to be reached beyond which it declines. At least this is true unless the first two leaves on the branch are to be considered as quite abnormal.

6. Nitrates. The figures given are calculated as Potassium Nitrate in the dry matter.

	Morning 7—8 A.M.	Noon	Evening 5—6 P.M.
	A. *Leaves (two) nearest the main vine.*		
	%	%	%
1st set	1·4	1·5	2·6
2nd „	1·5	1·7	1·3
3rd „	0·6*	1·2	1·4
4th „	1·1	1·4	1·4
5th „	1·1	2·5	1·4
	B. *Leaves (two) further from the main vine.*		
1st set	1·4	1·4	2·5
2nd „	1·5	1·9	1·5
3rd „	1·1	1·5	1·8
4th „	1·3	1·6	1·4
5th „	1·6	2·4	1·7
	C. *Leaves (two) furthest from the main vine.*		
5th set	1·5	2·1	1·9
	D. *Leaves from the main vine.*		
4th set	1·3	1·3	0·9

* This figure seems abnormal, and has been excluded from the averages.

Let us consider the variation of this very interesting constituent during he day, during development, and as due to the position of the leaf on the vine.

(a) *Variation of nitrates during the day.*

	Leaves nearest vine	Leaves further from vine	Leaves furthest from vine	Leaves on main vine
			(One set only)	(One set only)
	%	%	%	%
Morning	1·3	1·4	1·5	1·3
Noon	1·7	1·8	2·1	1·3
Evening	1·6	1·8	1·9	0·9

The amount of nitrates, except in the leaves on the main vine, seems to increase to a maximum in the day, and to remain about at this maximum till evening. The third set is an exception, and in both cases in this set the amount increases towards evening.

(b) *Variation of nitrates in similar leaves during development.*

	Leaves nearest vine	Leaves further from vine
	%	%
1st and 2nd sets (average)	1·7	2·0
3rd set	1·3	1·5
4th „	1·3	1·4
5th „	1·7	1·9

It seems quite clear from this that the variation in the amount of nitrates is not due to the stage of development, but either to the daily variation in the mechanism of absorption or to some other casual and local factor which our experiments do not reveal.

(c) *Variation of nitrates due to position on vine.*

	Leaves nearest vine	Leaves further from vine	Leaves furthest from vine	Leaves on main vine
	%	%	%	%
4th set	1·3	1·4	..	1·2
5th „	1·7	1·9	1·8	..

The position of the leaves on the branch seems to have very little effect on the quantity of the nitrate at any time. This is in accordance with our previous conclusions (see previous Memoir *loc. cit.*, page 20).

7. **Acidity.** The figures are reported as cubic centimeters of decinormal caustic potash required to neutralise the solution from 100 grammes of dry leaves.

	Morning 7—8 A.M.	Noon	Evening 5—6 P.M.
	A. Leaves (two) nearest the main vine.		
	%	%	%
1st set	95·8	109·1	86·0
2nd "	90·4	94·0	118·6
3rd "	108·4	71·6	99·5
4th "	126·5	70·3	72·3
5th "	126·8	70·3	70·0
	B. Leaves (two) further from the main vine.		
1st set	129·4	132·5	115·6
2nd "	114·6	96·6	116·2
3rd "	123·9	110·4	150·0
4th "	145·8	93·0	120·9
5th "	123·2	75·8	82·7
	C. Leaves (two) furthest from the main vine.		
5th set	114·6	80·9	106·5
	D. Leaves on the main vine.		
4th set	130·1	91·2	94·7

These results seem wholly irregular and do not indicate any constant change in acidity at different times of the day, or at different stages of growth as far as they are included in these series. The acidity, too, is not widely different from that recorded for the slightly over-mature leaves recorded on page 283.

We are now in a position to indicate in some measure what is the composition, in respect to the constituents we have determined, which is desired in leaves for chewing. These, it must be remembered, are usually mature or over-mature leaves, which have actually ceased growing, but have not become brittle, and are most often gathered in the morning. Those which are nearest to the main vine are usually disliked partly because their shape is not so regular, and partly also because they tend to be brittle, and also not to bleach well. Keeping this in mind we may say that, in leaves which are not yet mature, the starch and the non-reducing sugars rise during the day consistently and nitrates appear to do so as well, the reducing sugars (which form two-thirds of the total sugars present) remain fairly constant through the day as does also the tannin and the essential oil. In over-mature leaves, the proportion of reducing sugars is very much smaller, and they are not more than one-third of the total sugar present at any time, and the non-reducing sugars

are fairly constant and are present in by far the greater amount. The starch rises during the day, but slightly only. There is a slight increase in the tannin during the day, while the nitrates decline. The essential oil tends to rise at midday and then fall in the later part of the day.

The leaves, young and growing, and not yet sufficiently mature for eating, contain much reducing sugar, and little cane sugar. The sugar, as a whole, reaches a maximum and then with the maturity of the leaf appears to decline to a practically constant quantity. The same thing happens in the case of starch, and also, curiously enough, in that of tannin. After maturity is reached the tendency for both these is to fall. The essential oil rises to maturity and probably considerably beyond, but falls again when the leaf is considerably over-mature. The nitrates are fairly constant in quantity in similar leaves, throughout the life of the plant.

What is apparently wanted for chewing is a leaf with little starch and reducing sugar, and with an increasing quantity of cane sugar. Changes in the quantity of tannin do not seem material, but excess of this constituent is evidently not required. The value of the leaf is not conditioned by changes in the quantity of nitrates. The vital factor is, as we concluded from our previous paper, the amount of essential oil, which increases very largely as the leaf becomes more suitable for use, and which again declines when the leaf becomes over-mature. It is evident that it is in the study of this constituent, and the manner in which its quantity and character can be modified, that the investigation of the improvement of the leaf can be most profitably pursued.

II.

THE VARIETIES OF BETEL-VINE LEAF, THEIR QUALITIES, AND THE ESSENTIAL OIL IN THEM.

There are few products whose varieties are more widely distinguished by its users than the leaves of the betel-vine. Some, with special qualities, are associated with special places, and we have obtained leaf from a number of these centres, and have distilled the essential oil from most of them. We will now give an account of each of the kinds of leaf we have studied, and afterwards of the differences in their essential oil.

Poona green leaves. This variety is grown generally in the Deccan, and specially in Poona. In Bombay the leaves are much valued and used. They

are quite soft and sufficiently pungent to be relished both by occasional *pan*-eaters and by those addicted to *pan*-chewing. They bleach very well, and the well bleached leaves are said to be as good as the Ramtek variety.

Ramtek kapuri leaves. These are very soft, and form a good pulp in the mouth. They have a very faint camphor-like smell. Even if eaten to excess they have the reputation of not producing eruptions in the mouth as do many other varieties. They bleach well, and carefully managed, they will keep well. They are not so good in appearance as Poona leaves, and are large in size. In their own district, near Nagpur, they are the type most valued, as well as in other parts of the Central Provinces and Berar.

Ramtek bangla leaves. These, on the other hand, are not highly valued except by those who are strongly addicted to the habit of *pan*-chewing. They are considerably used by Mahomedans. They are very pungent, and have a very peculiar smell, difficult to describe, when chewed. They do not easily bleach, and are not used for this purpose. They have a local reputation at and near Nagpur.

Baroda bangla leaves. These are very similar in appearance and effect to the last described, and are likewise very pungent and much used by Mahomedans and by the so-called lower classes.

Ranibennur green leaves. Ranibennur is a great *pan*-growing centre in the Bombay Karnatak, and gives two types of leaf, the green and the black. The green leaves are eaten by the higher classes, and scarcely by others or by tobacco chewers. There is little local demand, most being sent to Bombay direct. They are not bleached locally, but in Bombay. They are valued for their softness, easy and good bleaching, good shape, and very good taste.

Ranibennur black leaves. These are used chiefly locally by the so-called lower classes and by those who also chew tobacco leaf. There is no export of these leaves to Bombay. They are very pungent, and are never bleached.

Chikodi green leaves. These leaves, grown in a number of centres in the Belgaum district, are very nearly identical with the Ranibennur green leaves, and very similar to the Poona green leaves.

In quoting these types we have naturally only dealt with those accessible to us. All, except the Ramtek leaves which are famous throughout India, are from the Bombay Presidency, but probably a study of these will give an idea of the nature of the differences in leaves from other places. The attached table is a summary of the qualities of the types we have examined.

Kind of Leaf	Average value per 1,000	Market	"Feel" of Leaves	Colour	Smell	Taste	Shape and Size
Poona green leaves (fresh) ...	Re. 0 12 0 to Rs. 1 4 0	Poona and Bombay.	Soft, not brittle nor tough.	Light green to green.	Nothing peculiar.	Pungent.	Well and regularly marked lines and generally beautiful.
Poona green leaves (bleached) ..	Rs. 2 0 0 to Rs. 3 0 0	Do.	Softer, do.	Yellowish white to yellow.	Do.	Slightly pungent.	Do.
Ramtek Kapuri green leaves (fresh) ..	Re. 1 0 0 to Rs. 1 8 0	Central Provinces and Berar.	Very soft, do.	Light green.	Camphor-like smell when chewed.	Do.	Flat, round, rather big.
Ramtek Kapuri green leaves (bleached) ..	Rs. 1 8 0 to Rs. 3 0 0	Do.	Even softer than the last.	Yellowish white.	Do.	Very slightly pungent.	Do.
Ramtek Bangla leaves ..	Re. 0 12 0 to Rs. 1 4 0	Nagpur and Ramtek.	Not soft, rather brittle and tough.	Dark green.	Very peculiar and strong smell when chewed.	Very pungent.	Small, ovate, rough.
Ranibennur green leaves ..	Re. 0 10 0 to Re. 1 0 0	Bombay and Locally.	Soft, not brittle nor tough.	Light green.	Nothing peculiar.	Pungent.	Like Poona.
Ranibennur black leaves ..	Re. 0 12 0 to Rs. 1 8 0	Locally.	Not soft, brittle and tough.	Very dark green.	Very pungent smell when chewed.	Very pungent.	Ovate, not big.
Baroda Bangla leaves ..	Re. 0 10 0 to Re. 1 0 0	Locally.	Not soft, brittle.	Dark green.	Do.	Do.	Round, not very small.
Chikodi green leaves ...	Re. 0 10 0 to Re. 1 0 0	Bombay and Locally.	Soft, not brittle nor tough.	Light green.	Nothing peculiar.	Fairly pungent	Like Poona.

We may, in fact, class these in several groups, as follows, leaving out the bleached leaves :—

1. (*a*) Poona green leaves.
   (*b*) Ranibennur green leaves.
   (*c*) Chikodi green leaves.
2. (*a*) Ramtek *kapuri* green leaves.
3. (*a*) Ramtek *bangla* green leaves.
   (*b*) Baroda *bangla* green leaves.
4. (*a*) Ranibennur black leaves.

In our opinion, it is probable that these are really two varieties represented respectively by (1) and (2), and (3) and (4) the former being green varieties, the latter dark green varieties.

From each of the types quoted in the above table, the essential oil was distilled with steam and the quantity determined, previous to our examination of the properties. The quantities obtained from market samples at the same time of the year are as shown below. Many determinations were made from Poona green and Poona bleached leaves and the figures for these give simply the limits we found.

	Essential oil per cent.
Poona green leaves	1·0 to 2·0
Ranibennur green leaves	1·8
Chikodi green leaves	2·7
Poona bleached leaves	2·0 to 4·0
Ramtek *kapuri* green leaves	2·0
Ramtek *kapuri* bleached leaves	3·1
Ramtek *bangla* green leaves	1·7
Baroda *bangla* green leaves	3·1
Ranibennur black leaves	3·1

It is, at once, evident from these figures that it is not merely the *quantity* of the essential oil which determines the value of the leaves for chewing, as the least valued of the leaves in some cases contain more essential oil than the most valued. If we except the Chikodi green leaves, none of the more valued light green leaves contain more than two per cent., unless bleached : on the other hand the less valued *bangla* and black leaves contain an average of 2·6 per cent. It was necessary, therefore, not only to consider the quantity of the essential oil but also its composition. This led to a study of the chemistry of *pan* oil.

III.

THE CHEMISTRY OF THE ESSENTIAL OIL OF BETEL-VINE LEAVES.

The work which has hitherto been done on the chemistry of betel-vine essential oil is well summarised in Gildemeister and Hoffmann's work on the Volatile Oils, and we may quote their description in part.

"When and by whom betel oil was first distilled is not known, it is highly probable that the preparation of the oil by Kemp[1] in 1885 was not the first. A superficial examination of Siam betel oil in the laboratory of Schimmel & Co.[2] in 1887 revealed the presence of a phenol in fraction 250—260°, which seemed to correspond with eugenol. Eykman,[3] who in 1888 examined an oil distilled by himself in Java, found no eugenol, but a new phenol which he called chavicol. A second examination in the laboratory of Schimmel & Co. revealed the fact that Siam betel oil contains neither eugenol nor chavicol, but a third phenol: a previously unknown isomer of eugenol, which Bertram and Gildemeister[4] in 1889 termed betel phenol.

"Betel oil is a light yellow to dark brown liquid of aromatic somewhat creosote-like odour, reminding of tea, and with a pungent taste.

"The specific gravity varies between 0·958 and 1·044: the oil from fresh leaves being lighter both in weight and colour than that distilled from the dried material. The rotatory power was observed on three samples of oil from fresh leaves. Of these two were lævogyrate (a_D up to $-1° 45'$) and one dextrogyrate ($a_D = +2° 45'$).

"With ferric chloride the alcoholic solution of betel oil produces a greenish to bluish-green colour."

The general result of the work which has been done on the chemistry of the essential oil shows that this oil consists of two groups of constituents, the first class consisting of one or more phenols of the class of eugenol or its isomers and analogues, and the second of a group of sesqui-terpenes of which cadinene was said to be isolated by Bertram and Gildemeister from Siam oil (*loc. cit.*).

Nearly all the work done (except that of Eykman) has been done in Europe on dried leaves—and all except the original preparation of Kemp, on

[1] *Pharmacographica Indica.*
[2] Bericht von Schimmel & Co., 1887.
[3] *Chemiker-Zeitung.* Vol. 12, page 1338.
[4] *Journal fr. Prakt. Chemie* (II), 39, p. 349.

Java or Siam oils. Our results differ considerably from those published hitherto, and we will simply indicate those which we have obtained.

Separation of Phenols and Non-Phenols in the oil. The phenols and non-phenols were separated by simply shaking the oil with a solution of caustic soda and measuring the loss in volume of the oil. In some cases 5 per cent. and in other cases 15 per cent. caustic soda was used. The percentage of matter dissolved with the latter was higher, while the former seemed to dissolve all the phenolic matter. The figures obtained with the two solutions were as follows with oil from bleached and unbleached leaves:

	5 % Soda Per cent. Phenols	15 % Soda Per cent. Phenols
Sample 1 (Green leaves)	41·0	46·0
Sample 2 (Green leaves)	42·0	49·0
Sample 3 (Bleached leaves)	55·5	64·0
Sample 4 (Bleached leaves)	56·0	68·0

In some cases, for comparison, a direct determination of the non-phenols was also made by extracting the mixed potash and oil repeatedly with ether, and evaporating the solution. This invariably gave a higher percentage of phenols than when the determination was simply made by loss of volume. The relationship between the figures is shown in the following table in five separate lots of leaf :—

	Phenols % (by direct reading of loss in volume)	Phenols % (by ether extraction)
Lot 1	70·0	75·3
Lot 2	82·0	90·0
Lot 3	56·0	73·2
Lot 4	45·0	61·2
Lot 5	65·0	66·5

In most of our determinations we have simply taken the direct reading of loss in volume, and the exact method of conducting the assay was as follows :—A burette of sixty cubic centimetres capacity calibrated in tenths of a cubic centimetre was almost completely filled with 5 per cent. caustic soda solution, and ten cubic centimetres of the oil added. The burette was stoppered, well shaken, and set aside for twelve to twenty-four hours. Drops of oil which might be adhering to the sides of the burette were then loosened by tapping or rotating the burette, and after the alkaline liquid had become clear the amount of non-phenol oil was read off.

Treated in this way, the various leaves described in the previous section gave figures as follows :—

	Phenols %	Character of the soda solution
Poona green leaves	42·0	Yellow coloured liquid, gradually becoming red.
Ranibennur green leaves	56·0	Dark coloured liquid.
Chikodi green leaves	65·0	Red coloured liquid.
Ramtek *kapuri* green leaves	70·0	Reddish coloured liquid.
Ramtek *bangla* green leaves	40·0	Very dark liquid.
Baroda *bangla* green leaves	45·0	Very dark liquid.
Ranibennur black leaves	39·0	Dark liquid.
Poona bleached leaves	56·0	Red coloured liquid.
Ramtek bleached leaves	82·0	Dark coloured liquid.

From these figures it is obvious that the percentage of phenols in the oil varies very much, and that the higher quality leaf contains the largest quantity of this class of substance, while the lower quality *bangla* or 'black' leaves contain considerably less. Further the effect of bleaching is to increase the percentage of phenols in the oil. Thus we have as follows :—

	Percentage of phenols	Increase by bleaching %
Poona green leaves	42·0	33
Poona bleached leaves	56·0	
Ramtek green leaves	70·0	17
Ramtek bleached leaves	82·0	

Nature of the Phenols in betel-leaf oil. On the question of the nature of the phenols contained in the oil, the information at present is about as contradictory as it possibly will be. As already stated (see page 299), it was first said to be eugenol : then no eugenol was found, and a new phenol named chavicol was obtained from Java oil : then Siam oil was found to contain neither chavicol nor eugenol, but another isomer of eugenol to which the name betel-phenol was given. None of these studies were made with Indian betel-leaf oil.

The three phenols mentioned are said to have the following properties.

Eugenol or allyl guaiacol [$C_6H_3 \cdot C_3H_5(1) \cdot OCH_3(4) \cdot OH(3)$] is the characteristic phenol of clove oil, and is also contained in cinnamon, sassafras, bay, and a number of other similar essential-oil-yielding plants. It is a yellow liquid, smelling of cloves, with a burning taste, boiling at 252°C. at 749 mm. *With ferric chloride in alcoholic solution it gives a blue colour. After shaking with water, the water solution gives no reaction with ferric chloride.* On oxidation it yields vanillin. *It forms a benzoyl compound with benzoyl chloride, melting*

at 69—70°C. and also gives an acetyl compound with acetyl chloride melting at 30—31°C.

Chavicol (*Para-allyl-phenol*) [C_6H_4. $C_3H_5(1)$, OH(4)], is said to occur in bay oil. It is a colourless liquid boiling at about 237°C. *Its aqueous solution is coloured intensely blue with ferric chloride, but the alcoholic solution only faintly blue.* Its benzoyl ester crystallises in long needles and melts at 72—73°C.[1]

Betel phenol. (*Chavibetol*) is an isomer of eugenol with the formula C_6H_3. $C_3H_5(1)$, $OCH_3(3)$. OH(4). It is not known from other sources than betel-leaf oil. It is a liquid with a smell much more like betel leaf than eugenol. It boils at 254—255°C. *It gives no colour with ferric chloride in aqueous solution, but an intensely bluish green colour in alcoholic solution. It gives a compound on treatment with benzoyl chloride, melting at 49—50°C. and crystallising in plates.* Acetyl chloride produces a compound boiling at 275—277°C. and melting at 5°C.

These three phenols can evidently be distinguished easily by (1) their reaction with ferric chloride in water and alcohol solutions, (2) by the melting point of their benzoyl compounds, (3) by the boiling point of the phenol. Investigation of the phenols from betel-leaf oil was made in these three directions. Most of the work was done on oil from Poona leaves which had not been dried.

Boiling point of the phenols from betel-leaf oil. The phenols for this inquiry were obtained by the method above described of treating the oil with 5 per cent. caustic soda, and then adding dilute sulphuric acid to the caustic soda solution. The phenols separated easily, and were then used for further experiments.

On heating the phenols thus obtained (which formed a dark red liquid), slight ebullition was noticed when the thermometer in the vapour reached 160°C. but no distillate came over. The thermometer rose then to 251°C.[2] and the liquid began to distil, and remained constant between 251—253°C. while the greater part of the liquid came over. The distillate was light green in colour.

In a second trial with a new lot of phenols, a small quantity distilled between 98°C. and 108°C. It had a peculiar smell rather like coriander : the

[1] The phenol which gave these figures for the benzoyl compound was prepared by Schimmel & Co., from oil distilled by De Vrij in Java. Its identity with chavicol is probable, but not proved.

[2] All these determinations of boiling points were made in Poona at an elevation of 1,800 ft. above sea level and are uncorrected.

quantity was very small, and it was chiefly water. No further distillate was obtained below 245°C. Between 245° and 253°C. the liquid distilled fast and the thermometer rose steadily without a break. Only a very small quantity remained at 253°C. when the temperature suddenly rose to 270°C., white fumes were evolved, and decomposition was evidently going on.

In a third trial with still a fresh lot of phenol, a very similar result was obtained at 105°C. Very nearly the whole of the remainder distilled between 246°C. and 251°C. the thermometer remaining constant at the latter figure. The distillate was very light green at first, but became reddish on standing.

This would seem to indicate that no considerable quantity of a substance with the boiling point of chavicol occurs in the Poona *pan* oil.

Reaction of the phenols with ferric chloride. The phenols were only slightly soluble in water. The water used for extraction when filtered gave a slight greenish blue colour with ferric chloride, which became intensely blue on addition of alcohol. This would again indicate that any substance having the properties of chavicol can only exist in traces. As water shaken with eugenol does not give any reaction with ferric chloride, there must be some quantity at least of another phenol probably betel-phenol.

Investigation of the benzoyl compounds of the phenols. The benzoyl compounds seemed to form the most likely means of ascertaining the nature of the phenols, and hence these were prepared from all the varieties of the oil in our hands.

About five grammes of oil from Poona leaves was taken in the first instance, and treated with 15 per cent. caustic soda solution (20 grammes), and 5 grammes of benzoyl chloride added the solution being vigorously stirred. An oily precipitate was formed, becoming semi-solid at 10°C. It was then dissolved in hot alcohol, and fractionally crystallised. A small residue was finally obtained from which crystals could not be obtained. The separate fractions crystallised as follows :—

	(1) Original crystals M. P.	(2) After recrystallisation M. P.
1st fraction	68°C.	68°C.
2nd ,, ...	67°C.	67°C.
3rd ,, ...	67°C.	67°C.
4th ,, ...	64°C. (yellow)	66°C.
5th ,, ...	67°C.	67°C.
6th ,, ...	67°C.	67°C.
7th ,, ...	51–53°C.	51–53°C.
8th ,, ...	50°C.	50°C.
9th ,, ...	49–50°C.	49–50°C.
10th ,, ...	Uncrystallisable	

All these tests point, therefore, to the fact that while the bulk of the phenols in the essential oil consist of eugenol, there is present with it another phenol having the properties of the betel-phenol previously described. This can be separated by washing the phenols with water, as it is much more soluble in water than the eugenol. This is shown by the following experiment.

10 grammes of the oil were treated with 5 per cent. caustic soda solution (100 c.c.) well shaken, and kept for a day. The non-phenolic matter was then removed, and traces remaining with the alkaline liquid taken away by washing the latter with ether. The residual liquid was treated with dilute sulphuric acid when the phenols separated as a black oily liquid, and the soluble portion was extracted by ether. The percentages obtained thus were as follows:

Non-phenols	29·45 % of the oil.
Total phenols	70·55 % of the oil.

The purified phenols were then shaken with water and filtered. The filtrate gave a slight greenish blue colour with ferric chloride, becoming intensely blue on addition of alcohol. The portion soluble in water was 5·71 per. cent of the original oil.

The separated water-soluble phenol was treated with benzoyl chloride to obtain the benzoyl derivative—and the product crystallised. Crystallisation was not satisfactory, however, either from alcohol, acetic ether or any other liquid tried, even in a freezing mixture. A hard mass was left on evaporation of the solvent which melted at 51—52°C. A very very few crystals separated from alcoholic solution on keeping for a month, which melted at 63—64°C. and which were evidently the benzoyl compound of eugenol. Everything points to this water soluble phenol being a mixture of betel phenol with a small quantity of eugenol, and the two phenols can be partially separated by means of this difference in solubility.

As the determination of the exact nature of the phenols from the betel-leaf oil seemed a matter of importance, and the examination of the benzoyl compounds seemed to afford the most satisfactory means of making it out, a number of other experiments were made as follows :—

(1) *Oil from Poona green leaves.* The phenols and non-phenols were separated by means of 5 per cent. caustic soda solution, the solution treated with sulphuric acid, and the phenols separated from the solution completely by washing with ether. They were then treated in the usual way (vide *supra*) with benzoyl chloride, and the derivative washed with hot water and dissolved in warm alcohol. The solutions of the more easily and of the less easily soluble

portions were kept separately, and allowed to deposit crystals slowly. In both cases big crystals separated after ten days, and gave melting points as follows :—

(1) derivative from the most easily soluble phenols M. P. 66—67°C.

(2) derivative from least easily soluble phenols—M. P. 66°C.

No further crystals were obtained in this case.

(2) *Oil from Poona green leaves.* In this case the phenols and non-phenols were separated by 5 per cent. caustic soda as usual, and the caustic soda solution treated directly with benzoyl chloride. The benzoyl derivative was dissolved in alcohol in two fractions. The solution of the more easily soluble portion gave crystals melting at 66—67°C. ; that of the less easily soluble portion gave crystals melting at 61—66°C. On recrystallisation all melted at 66—67°C. The soluble on evaporation gave a residue not containing crystals and which we failed to get to crystallise.

(3) *Oil from Poona bleached leaves.* The experiment was carried out with this oil as in No. 1. All the crystals obtained melted at 66—67°C., but there was a residue in the liquid which we did not succeed in making crystallise.

(4) *Oil from Poona bleached leaves.* This experiment was carried out exactly as in No. 2, but all the crystals obtained melted at 66—67°C. The residual solution could not be made to produce crystals.

(5) *Oil from Poona green leaves.* The phenols were separated as usual with 5 per cent. caustic soda, followed by separation with sulphuric acid. The separated phenols were then repeatedly washed with cold water to get the portion soluble in water, and this water solution was then treated with benzoyl chloride to obtain the benzoyl derivative. This benzoyl derivative crystallised with very great difficulty. On evaporating the alcoholic solution a thick reddish semi-liquid residue was obtained which became hard and solid on rubbing with a glass rod. This hard substance melted at 50—51°C.

The mass was treated with a very small quantity of alcohol insufficient to dissolve the whole. The undissolved residue then melted at 50—52°C. : the solution deposited a very small quantity of crystals melting at 66°C. and a further evaporation gave a residue melting at 50—52°C.

The water soluble phenols in this case consist, therefore, chiefly of a phenol giving a benzoyl derivative melting at 50—52°C. mixed with a small quantity of a phenol whose benzoyl derivative melts at 66°C.

The result of all these experiments seems clear. Two phenols exist in the oil. These, judging by the benzoyl compounds, correspond with eugenol

and betel-phenol. The former is present in by far the largest amount, and is only very slightly soluble in water; the latter (betel-phenol) on the other hand is very fairly soluble in water, and remains in the water-soluble portion mixed with a little eugenol. The portion, from Poona green leaves, soluble in cold water was in one case 5·7 per cent. of the phenols. This will represent slightly more than the proportion of betel-phenol present.

The principal phenol present was always eugenol. In our samples this formed about ninety-five per cent. of the total.

Examination of the non-phenols in betel-leaf oil. The only other materials which have been isolated from betel-leaf oil are terpenes and sesquiterpenes. The position of our knowledge with regard to these is thus stated by Gildemeister and Hoffmann[1].

"Another constituent possibly to be found in all betel oils is cadinene $C_{15}H_{24}$. So far this hydrocarbon has been isolated from the Siam oil only (dihydrochloride M. P. 118°C.). Probably the sesquiterpene (B. P. about 260°C. Sp. gr. 0·917), found by Eykman in the betel oil from Java is also cadinene. The Java oil from fresh leaves contains a considerable amount of low boiling constituents. Eykman[2] did not succeed in isolating or identifying a pure terpene of constant boiling point from the fractions between 173–190°C. Probably several terpenes are present, but apparently no pinene. Fraction 173–175°C. (Sp. gr. 0·848 at 16°C. : α_D = – 5° 20′) yielded neither a solid bromide nor a crystalline hydrochloride. Fractions 190–220°C. contain substances having a minty odour (menthone or menthol ?).

"The oil distilled from dried Siam leaves was devoid of low boiling fractions, only a few drops coming over below 200°C."

We have not as yet done more than a preliminary examination of the non-phenolic portions of the essential oil, but the results so far obtained may be recorded.

The non-phenolic portions of the betel-leaf oil were distilled at ordinary pressure and gave fractions (Poona green leaf) as follows :—

No.			%
1	185–200°C.	Chiefly from 195 to 200°C.	7·4
2	210–230°C.	Chiefly from 220 to 230°C.	5·3
3	243–245°C.	Chiefly at 244°C.	6·5
4	247–250°C.	Chiefly at 250°C.	8·4
5	250–255°C.	Chiefly at 253–254°C.	29·2
6	256–263°C.	Chiefly at 257°C.	5·3

[1] *The Volatile Oils*, page 329.
[2] *Chem. Zeitung*. Volume 12, page 1338.

At 263°C. white fumes commenced to be evolved, and it was evident that decomposition was commencing. The distillation was therefore completed under reduced pressure (350 mm.) and the following further fraction was obtained.

No.		%
7	240–250°C. (at 350 mm. pressure)	11·1
Residue (black in colour)	.. ..	6·8
		100·0

Of these fractions the following notes were made :—

No.	
1	Colourless liquid with highly fragrant sweet smell.
2	Ditto ditto.
3	Slightly greenish liquid, with slight fragrant sweet smell.
4	Greenish liquid, only very slightly fragrant.
5	Liquid, more green than No. 4, with slightly objectionable smell.
6	Liquid, more green than No. 5, with very nasty smell.
7	Very green liquid.

The distillate No. 6 was redistilled and the green colour was found to be persistent in the distillate.

The specific gravity of each of the fractions was obtained as follows (at 25°C.):

No.	
1	0·885
2	0·905
3	0·944
4	0·951
5	0·952
6	0·952
7	0·960

We were unable to detect either cadinene or caryophyllene in any of the above fractions. The examination will however be continued.

Conclusions regarding the essential oil of betel-leaf. The essential oil of betel-leaf consists essentially of two portions, consisting respectively of phenols and of terpene-like bodies. The relative proportion of these varies, and the higher the quality of the leaf, the higher the proportion of phenols in the essential oil. The proportion of phenols in our samples varied from 12 per cent. (Poona) to 70 per cent. (Ramtek *kapuri*) in green leaves of the light green variety, and from 39 per cent. to 45 per cent. in green leaves of the dark green variety. The bleaching of the leaves not only increases very much the total quantity of the essential oil, but also the proportion of the phenols in it. In two cases where bleaching was carried out the increase in the percentage of phenols was from 17 to 33 per cent.

The phenols consist essentially of eugenol in all our cases, mixed with a nall percentage of betel phenol. The latter can be largely separated by ashing the phenols with water in which it is very much more soluble than ıgenol. No sign of any substance having the properties attributed to chavicol[1] ıs been found in any of our samples.

The non-phenolic portion of the essential oil is a mixture of a number of ıbstances as yet uninvestigated. Over 60 per cent. boils between 240° and 55°C. This has a light green colour, and a somewhat objectionable smell. ; is not cadinene or caryophyllene.

IV.

The Commercial Bleaching of the Betel-Vine Leaf.

In our former memoir we discussed the method by which the bleaching f betel-vine leaf is carried on, and found that, as normally conducted, the rocess results in a very large increase in the amount of essential oil present, ı a very considerable increase in the acidity, in a large disappearance of the tarch and non-reducing sugars, and in a large decrease in the amount of iastase. We found moreover that oxygen made the process more rapid, nd that its absence prevented the change going on. It appeared also that ight acidity was necessary to the process. The problem now before us was ɔ bleach the leaf more rapidly with equal production of essential oil and to btain a leaf which has the further qualities desired by the consumer.

As usually conducted the process of bleaching is briefly as follows. For ırther details we must refer the reader to our former memoir (*loc. cit.*).

About fifty to seventy pounds of leaves are packed together in a round asket two to two and a half feet in diameter, and twelve to eighteen nches high, which has been lined with matting. The method of packing ; first to cut off the stalks, and then arrange the leaves almost vertically, in ıyers, forming three or four circular rings of leaves in the basket in each ıyer. A vertical hole is reserved in the centre, of convenient diameter to llow the hand, when introduced, to reach the bottom layer easily. The vhole is then sprinkled with water, and covered with moist gunny cloth, so hat it remains in the dark. After three or four days the gunny bag is removed, nd the leaves examined. After eight days all are taken out, the bleached eaves and the rotten leaves removed, and the remainder rearranged. All eaves becoming turmeric yellow and not white, and all with red or brown atches are removed. This taking out and picking over is repeated every

five or six days, until practically all the leaves have become bleached, or rotten, or refuse to change colour.

Properly bleached leaves should be very soft, tender, of a sunflower yellow to whitish yellow colour, very bright, clear of any stain as if rotting had commenced, and should become brighter on keeping. A brittle leaf is not liked, and leaves too highly bleached are objected to. When well prepared, bleached leaves are worth three times as much as the original green leaves from which they are obtained.

As already stated, our former work proved that the process was one in which oxidation takes a prominent part, and that slight acidity is necessary to its successful carrying out. The experiments now to be recorded deal with

(*a*) the influence of light on the bleaching process,

(*b*) the influence of moisture on the bleaching process,

(*c*) the influence of varying temperature on the bleaching process.

We have been able to make distinct progress in ascertaining the conditions which lead to rapid and successful bleaching, with the minimum amount of loss on account of the rotting of the leaves.

(*a*) *Influence of Light on the Bleaching Process.*

It will be noticed from the above description that in the process in common use, the bleaching of *pan* leaf is carried out in the dark. The basket in which it takes place is always carefully covered, and only at the time at which the picking over of the leaf takes place is the material exposed to the light. The first experiments undertaken, therefore, were to ascertain whether this is a necessary precaution, or whether the presence of light is an indifferent matter.

The first experiment compared the following conditions :

(1) Green leaves dipped in water, the excess of water shaken off, and then kept in a nearly saturated atmosphere *exposed to light.*

(2) Green leaves kept in a saturated atmosphere, airtight, also *exposed to light.*

(3) Green leaves kept in a nearly saturated atmosphere, in a dark cupboard, and hence only exposed to light occasionally.

(4) Green leaves kept in a nearly saturated atmosphere, carefully protected from all access of light by means of a dark opaque cloth.

Leaves (eight or nine in each case) kept in these various conditions were watched and examined each day for twenty-four days. The results were clear. Whether judged by the amount of rotting which took place, by the rapidity,

or by the regularity of bleaching, it was evident that light was a distinct disadvantage to the process. The temperature throughout was 24° to 26°C.

In the matter of rotting, after the process was over we had as follows :—

	No. 1 Exposed to light	No. 2 Exposed to light and limited amount of air	No. 3 Kept in the dark	No. 4 Kept in complete darkness
	%	%	%	%
Bleached	None	12½	78	78
Partially bleached	22	12½	11	None
Rotting	45	37½	11	22
Will not bleach	33	37½	None	None

It is evident, therefore, that access of light almost entirely stopped the bleaching process.

Notes made of the process were as follows :—

No. 1 (*exposed to light*). The proper form of bleaching hardly commenced at all. A certain number of leaves began to turn yellow, and in this sense the process was rapid in the first instance. The yellowness, however, rapidly changed to rotting and eventually most of the leaves were no use either for eating or for further keeping.

No. 2 (*exposed to light, with limited amount of air*). The proper bleaching in this case also hardly began. The leaves only got yellow spots here and there on the surface. Futher bleaching hardly took place except in one case.

No. 3 (*kept in the dark*). The bleaching was slow, but it went on regularly and continuously. Of the bleached leaves, about half were bleached white and half yellow. The bleached leaves were lustrous, and satisfactory from a consumer's point of view.

No. 4 (*kept in complete darkness*). The leaves bleached slowly and normally, the majority tending to yellow rather than white, although the number of white-bleached leaves was thirty-three per cent. of the total taken.

A second experiment was now undertaken with a larger number of leaves, generally 64 or 65, weighing in each case one hundred grammes. We compared the following conditions :—

(1) Bleaching with exposure to light.
(2) Bleaching in the dark.
(3) Bleaching exposed to *blue* light.
(4) Bleaching exposed to *red* light.

The results confirmed the previous experiment, and in addition showed that the exposure to red light was much more fatal to satisfactory bleaching than exposure to blue light. The results tabulated, after twelve days, were as follows :—

	No. (1) Exposed to light	No. (2) Kept in the dark	No. (3) Exposed to blue light	No. (4) Exposed to red light
	%	%	%	%
Total bleached leaves	None	71	60	36
(a) White bleached	..	46	53	10
(b) Yellow bleached	..	25	7	26
Half bleached leaves	63	None	None	None
Total rotting leaves	37	29	40	64
(a) Leaves rotting while green ..	37	None	5	34
(b) Leaves rotting after partial bleaching	..	29	35	30

Thus the maximum amount of rotting took place under the red light, and that under the blue and the white light was about equal. The least took place in the dark. The maximum amount of bleaching took place again when the leaves were kept in the dark. The proportion of the bleached leaves which became white and yellow respectively was

	No. 1	No. 2	No. 3	No. 4
	%	%	%	%
White bleached	None	65	89	29
Yellow bleached	None	35	11	71

The yellow bleached leaves are, of course, considered far inferior to those which are white.

(b) *Influence of Moisture on the Bleaching Process.*

It was noticed in the experiments on the action of light that if the leaves became in the slightest degree dry at the edges or elsewhere, rotting set in almost immediately. Experiments were undertaken, therefore, to ascertain the effect of a perfectly saturated atmosphere or other conditions of moisture on the bleaching process.

The conditions compared were the following :—

1. The leaves were placed in an atmosphere kept nearly saturated by being surrounded with wet gunny cloth, and placed under a bell jar. The gunny cloth did not, however, touch the leaves.

2. The leaves were placed in a similar position to the last, except that the bell jar itself was standing in water. The gunny cloth did not touch the

leaves. The atmosphere may be considered to have been a little more nearly saturated than in the last case.

3. The leaves were placed in a similar position to No. 2, but the wet gunny was re-wetted every morning, and so the conditions were more nearly saturated with moisture than even in the last case.

4. The leaves were placed under similar conditions to No. 3, but the end of the gunny cloth itself was continually dipping in water, and so it was kept continuously re-wetted.

5. The leaves were placed under similar conditions to the last, except that they were surrounded by wet leaves (as they would be in the regular process) which were not included in the experiment.

The leaves kept in No. 1 were, therefore, in the conditions of least saturation, and the saturation was greater in each succeeding case.

The leaves taken in each case weighed one hundred grammes and were from 54 to 60 in number, and the general results were as follows :—

	No. 1	No. 2	No. 3	No. 4	No. 5
	%	%	%	%	%
Total bleached leaves	52	81	70	54	47
(*a*) White bleached	44	58	64	35	41
(*b*) Yellow bleached	8	23	6	19	6
Total rotting leaves	48	19	30	46	53
(*a*) Rotting while green ..	21	10	3	13	12
(*b*) Rotting after partial bleaching	27	9	27	33	41

Of the bleached leaves, the percentage bleached white and yellow was as follows :—

	No. 1	No. 2	No. 3	No. 4	No. 5
	%	%	%	%	%
White bleached leaves	84	72	92	65	87
Yellow bleached leaves ..	16	28	8	35	13

The results thus shown seem to indicate that while enough moisture must be present to ensure that not the slightest drying should take place, more than this is injurious. The best results were obtained (Nos. 2 and 3) when every precaution was taken to ensure that the atmosphere was nearly saturated with water, but when the leaves themselves were not wetted, except at the beginning. If the atmosphere was more nearly saturated than was required to keep the leaves from drying, the amount of rotting rapidly increased, and the number of leaves bleaching properly became very much reduced.

In No. 1, there was a slight sign of the leaves drying at the edges : this has probably caused the very inferior results obtained in that case, and this accounts for the large number of leaves which rotted before bleaching took place.

(c) Influence of Temperature on the Bleaching Process.

It had now become clear that for correct bleaching to occur the leaves must be in the dark, and in an atmosphere nearly enough saturated to prevent the slightest drying. The next point to ascertain was the temperature at which the best results were obtained.

The temperature was obviously an important factor in the process. In the bleaching of *pan* leaves, as commercially carried on, the results are recognised to be much better during some seasons than during others. Thus, for instance, the period from February to May in Poona is the best time for making good bleached leaves, and this corresponds roughly to the hot weather in this part of the country. This is partly due also to the fact that the *junawan* leaves, which give the best results, are more readily obtainable at this period of the year, but the temperature must be recognised to be a factor in the operation.

We, therefore, carried on a number of experiments on this point. In the first of these the process of bleaching was carried on respectively at the room temperature ranging from 26° to 30°C, in an incubator kept at 28° to 31°C, and in an incubator at the higher temperature of 31° to 33°C. The difference between these temperatures is not great, but it is sufficient to have a considerable influence on the result.

The result of the bleaching in each case was as follows, the number of leaves taken being from 62 to 66 :—

	No. 1 26° C. to 30° C.	No. 2 28° C. to 31° C.	No. 3 31° C. to 33° C
	%	%	%
Total bleached leaves	47	64	63
(a) White bleached	41	37	35
(b) Yellow bleached	6	27	28
Total rotting leaves	53	36	37
(a) Rotting while green	12	None	None
(b) Rotting after partial bleaching ..	41	36	37

The result at 26° to 30°C. was obviously considerably below the average, but the other two, on similar leaves, were practically equal and were in each case superior to the average, except that the proportion of yellow bleached leaves was higher. But during the process it was noticed that at the higher

temperature the leaves bleached (1) much more quickly, and (2) much more evenly than under ordinary room conditions. The difference in time was considerable. While the bleaching was not completed in No. 1 (the room temperature) until after twelve days, in Nos. 2 and 3 the process was finished after five days. This difference in time probably accounted for the smaller amount of rotting, and also for the rotten leaves rotting after partial bleaching rather than before.

This result was so promising that further experiments were undertaken with larger quantities of leaf. In the first of these we compared :

(1) the bleaching of the leaf under the ordinary room conditions and by the usual method adopted by the *tambolis* (temperature 27°C. to 28°C.)

(2) the bleaching of the leaf at a higher temperature in an incubator (temperature 30° to 35°C.)

(3) the bleaching of the leaf at a much lower temperature, in a box surrounded by ice (temperature 8° to 16°C.).

It may at once be said that no real bleaching took place at the lower temperature (8° to 16°C.). The leaves gradually lost their lustre, becoming dark in colour, and showed signs of decay by the fifth day. This darkening and decaying continued and after a few days further the whole lot had to be thrown away.

In the other two cases the following table shows the general results in the first case for an ordinary bazaar sample of leaves, in the second for a selected sample of *junawan* leaves.

A.—*Bazaar leaves (without selection).*

	No. 1 27°–28° C.	No 2 30°–35° C.
Time required for bleaching	17 days	10 days
	%	%
Total bleached leaves	**35**	**84**
(*a*) **White bleached**	**9**	**68**
(*b*) **Yellow bleached**	**18**	**14**
(*c*) **Partially bleached**	**8**	**2**
Total rotting leaves	**65**	**16**
(*a*) **Rotting while green**	**21**	**4**
(*b*) **Rotting after partial bleaching**	**34**	**12**

B.—*Junawan leaves.*

Time required for bleaching	No. 1 27°–28° C. 21 days	No. 2 30°–34° C. 9 days
	%	%
Total bleached leaves	74	83
(a) White bleached	30	42
(b) Yellow bleached	35	41
(c) Partly bleached	9	..
Total rotting leaves	26	17
(a) Rotting while green	10	1
(b) Rotting after partial bleaching..	16	16

It is evident from these figures, therefore, that the increased temperature has not only increased the speed of the bleaching but it has made it possible to bleach leaves, which are not usually considered fit for this purpose, almost equally well with those which are specially selected for it. Further, even among leaves considered as particularly suitable, a larger percentage of bleached leaves has been obtained at the higher temperature, and less rotting than is normally the case.

The bleached leaves at the higher temperature were of a better colour and more evenly bleached appearance than those conducted by the ordinary process at the ordinary room temperature, and nearly all the leaves were ready on the fifth or sixth day of the process.

It was incidentally noticed during these experiments that leaves whose edges touched metal almost inevitably went rotten, and the same was the case to a less extent if they were in contact with gunny. Loss was minimised if they were in contact with dry plantain leaves, wetted for the occasion.

An attempt was now made to conduct the process at a still higher temperature, namely, 36° to 38°C. but the process totally failed. The leaves began to show signs of drying on the third day, in spite of the saturation of the atmosphere—yellow patches then appeared on all the leaves and these were quickly followed by a general appearance of cooking, and final darkening in colour and decay.

So far the results were obtained with either ordinary leaves or *junawan* leaves. The younger leaves from the betel vines, notoriously unsuitable for

bleaching by the ordinary process usually called *navati* leaves—were now taken in order to see how their bleaching would be affected by raising the temperature. The results of an experiment similar to that described for *junawan* leaves were as follows :—

A.—*Ordinary navati leaves.*

	No. 1. 24° to 27° C.	No. 2. 29° to 33° C.
Time required for bleaching	26 days	9 days
	%	%
Total bleached leaves	32	75
(*a*) White bleached	11	15
(*b*) Yellow bleached	None	60
(*c*) Partially bleached	21	None
Total rotting leaves	68	25
(*a*) Rotting while green	45	16
(*b*) Rotting after partial bleaching	23	9

B.—*Selected navati leaves.*

	No. 1. 24° to 27° C.	No. 2. 25° to 28° C.	No. 3. 29° to 33° C.
Time required for bleaching	24 days	18 days	9 days
	%	%	%
Total bleached leaves	65	58	73
(*a*) White bleached	7	26	31
(*b*) Yellow bleached	24	32	42
(*c*) Partially bleached	34*	None	None
Total rotting leaves	35	42	27
(*a*) Rotting while green	18	15	8
(*b*) Rotting after partial bleaching ..	17	27	19

* A large number of these leaves showed signs of rotting if further bleaching was attempted. They would not have bleached if kept longer.

The result here is remarkable. By raising the temperature to 29° to 33°C. the *navati* leaves, usually considered as unsuitable for bleaching, have given nearly as good results as the *junawan* leaves so far as total bleaching is concerned, though the proportion of bleaching 'yellow' is greater. Similar results have been obtained in repeated trials, and it may be taken that if the process be conducted at the higher temperature, a much larger range of leaves may be used for the preparation of the bleached product than would otherwise be suitable.

The limits of temperature between which the bleaching process is practicable seem, in fact, fairly well defined. We have never conducted a successful

bleaching below 24° to 25°C., with any kind of leaves, although the leaves do actually bleach slowly as low as 20°C. The upper limit seems to be 33°C. for ordinary or *navati* leaves, and 34° to 35°C. for the best *junawan* leaves. With *navati* (younger) leaves, even 33°C. seems a little too high. As the upper limit is approached, the operation becomes more rapid and regular, but above the point indicated the process will not work, practically, at all.

When the leaves are nearly bleached, it also appeared that the temperature should be lower than in the early stages. If such leaves are suddenly put into a chamber at 32°C., they begin to turn yellow and then suddenly to start rotting. When such leaves are kept at the ordinary temperature (that is to say, from 25°C. to 26°C.), they remain in good condition without much loss in lustre or colour for a long period.

The actual loss in keeping well-bleached leaves at 25° to 26°C. was tested with forty-five leaves, kept in an atmosphere nearly enough saturated to prevent drying. All were rotted after a month, and the progress of the decay is shown in the following table :—

Days	Number of leaves good	Number of leaves rotting	Percentage of leaves rotting
Beginning	45	..	..
After 15 days	30	15	33·3
After 25 days	25	20	44·4
After 27 days	14	31	68·9
After 29 days	7	38	84·4
After 31 days	None	45	100·0

So far we have only considered the effect of temperature on the bleaching of the leaves as judged by appearance. It has been previously indicated, however, that the process derives its value largely from the fact that, as usually conducted, it is accompanied by an increase in essential oil, and especially in the phenolic portion of the essential oil. We have now to consider whether conducting the process at a temperature higher than that usual will lead to an equal increase in essential oil. That is to say :—Are the change in colour and the increase in essential oil parts of one process,—or are they independent changes going on in the leaf ? In the former case, the change in colour would be all we need aim at : in the latter it might sometimes be an indication of the highest quality of bleaching, and sometimes not. Experiments were undertaken to test this point.

At first a few leaves only were kept under various bleaching conditions, and the essential oil determined in each case from those which bleached in a more or less satisfactory manner. The results were as follows :—

A.—*Bazaar sample of junawan leaves.*

	Essential oil Per cent.
Fresh green leaves	2·08
Leaves bleached by usual method	3·82 (greenish white).
Leaves bleached at 30° to 32° C.	2·64 (very thoroughly bleached).
Leaves kept for bleaching below 20°C.	1·72 (somewhat yellow).

B.—*Selected sample of junawan leaves.*

	Essential Oil Per cent.
Fresh green leaves	4·31
Leaves bleached by usual method	6·17 (yellowish white).
Leaves bleached at 30° to 32° C.	8·42 (white).
Leaves bleached at 32° to 35° C.	3·84 (yellow).

It seemed probable, therefore, that the process of bleaching and the production of essential oil were distinct processes, that the production of yellow leaves was not associated with an increase in essential oil while that of white leaves was, and that if the temperature be increased too far, the essential oil does not increase at all.

A larger lot of leaves was taken, under more normal bleaching conditions than could be given in a small sample, and the effect on the essential oils determined.

A.—*Bazaar sample of junawan leaves.*

	Essential Oil Per cent.
Fresh green leaves	2·15
Leaves bleached by usual method (at 27° to 28° C.) ..	3·14 (greenish white).
Leaves bleached at 30° to 34° C.	2·64 (very thoroughly bleached).

B.—*Selected sample of junawan leaves.*

	Essential Oil Per cent.
Fresh green leaves	4·58
Leaves bleached by usual method (at 27° to 28° C.) ..	5·14 (greenish white).
Leaves bleached at 30° to 34° C.	4·51 (very thoroughly bleached).

At a temperature of 30° to 34°C., therefore, while the bleaching was excellent from a point of view of appearance, there did not seem in this experiment to be the same increase in essential oil as was obtained under the more usual conditions. This proves, we think, that the processes of change of colour,

and of increase in essential oil are largely independent—but it is still a question whether the higher temperature causes less production of essential oil, or whether under the very thorough bleaching, usually obtained, it is first formed and afterwards disappears.

That the latter is probably the case was indicated by an experiment with ordinary *navati* leaves, in which the bleaching was not allowed to go so far, and which gave the following results :—

Ordinary navati leaves.

	Essential Oil Per cent.
Fresh green leaves	1·36
Leaves bleached by usual method (at 24° to 27° C.) ..	2·83 (yellowish).
Leaves bleached at 29° to 33° C.	3·07 (yellowish white).

Here the leaves bleached at the higher temperature are fully as good as those bleached at 24° to 27°C., but with them the process has not been carried so far as in the cases previously given.

The question of the relationship of the change in colour, and the increase in essential oil could, however, only be settled by following, from day to day, the growth of essential oil in a batch of bleaching leaves, both under ordinary conditions and under conditions of higher temperature. This has been done with both ordinary and selected *navati* leaves.

A.—Ordinary navati leaves.

(1) Bleached at 27° to 28°C.

	Essential Oil Per cent.
Original leaves	2·27 (green).
Leaves four days later	2·67 (greenish).
Leaves six days later	2·70 (greenish yellow).
Leaves eight days later	2·41 (greenish yellow).
Leaves ten days later	2·81 (greenish yellow).
Leaves twelve days later	3·14 (yellowish white).
Leaves fourteen days later	2·47 (yellowish).

(2) Bleached at 29° to 33°C.

	Essential Oil Per cent.
Original leaves	2·27 (green).
Leaves two days later	2·02 (greenish).
Leaves four days later	3·09 (yellowish).
Leaves six days later	2·99 (yellowish white).
Leaves eight days later	1·83 (very thoroughly bleached).

B.—*Selected navati leaves.*

(1) Bleached at 27° to 28°C.

	Essential Oil Per cent.
Original leaves	2·01 (green).
Leaves two days later	2·31 (green).
Leaves four days later	2·36 (green).
Leaves six days later	2·44 (greenish).
Leaves eight days later	2·41 (greenish).
Leaves ten days later	2·74 (greenish yellowish).
Leaves twelve days later	2·46 (greenish yellowish).
Leaves fourteen days later	2·52 (greenish yellow).
Leaves sixteen days later	3·16 (greenish yellow).
Leaves eighteen days later (average)	3·40 (yellowish white).
Leaves eighteen days later (best bleached leaves, selected)	5.00 (yellowish white with green tinge).

(2) Bleached at 29° to 33°C.

	Essential Oil Per cent.
Original leaves	2·01 (green).
Leaves two days later	2·15 (greenish).
Leaves four days later	2·88 (greenish yellow).
Leaves six days later (average)	3·17 (yellowish white).
Leaves six days later (best bleached leaves, selected)	4·06 (yellowish white with green tinge).
Leaves eight days later	2·00 (very thoroughly bleached).

These figures show in every case that the increase in essential oil is a continuous process, until the leaves reach a yellowish white colour, with a tinge of green. If they are bleached further than this the amount of essential oils rapidly declines again. If this fact be taken into account the bleaching at a higher temperature gives almost, if not quite, as large an increase in essential oil as in the process as usually carried out—but the danger of over-bleaching in this case is considerably greater.

The process and its results, as conducted at a higher temperature, have been shown to some of the *tambolis*, who have highly approved of the resulting leaf, and of the reduced amount of rotting thus obtained. It remains, however, to devise an apparatus in consultation with the *tambolis*, which they can use in practice, in order to yield equally good results, and this will be the next step in bringing the results here indicated into use by the manufacturers of bleached betel vine leaves.

In the meantime, our results seem to prove .

(1) that the bleaching of betel vine leaf can only be carried out in the absence of light. Under white light little or no normal bleaching takes

place. Both red and blue lights are injurious, the former much more so than the latter ;

(2) the atmosphere under which bleaching is conducted must be sufficiently saturated to prevent the slightest drying of the leaves—as drying is always followed by rotting. Greater saturation than is required to keep the leaves from drying leads, however, to increase in the rotting ;

(3) the bleaching process can be carried on much more rapidly and evenly if the temperature be increased to 29° to 33°C. A temperature higher than 35°C. (or less with *navati* leaves) is fatal and yields no bleached leaves—and a temperature lower than 24°C. is equally useless ;

(4) bleaching at a higher temperature (29° to 33°C.) if checked at the proper point, leads to almost as great an increase in essential oil as if the bleaching is done at the ordinary temperature, as is usual—and as it gives less loss from rotting of the leaves, and can be carried on more rapidly, it has very considerable advantages. Nearly the same results can be obtained in six days as require sixteen days by the more usual process ;

(5) if bleaching is carried on beyond the stage when the leaves are yellowish white, the amount of essential oil rapidly declines, and hence the leaves should be removed at this stage. As the rapidity of the process is greater there is more danger of such over-bleaching when it is conducted at a higher temperature as here suggested ;

(6) it only now remains to devise an apparatus which the *tambolis*, who prepare the bleached leaves, can use, and this is now being done.

CONCLUSION.

Such are the results which we have now obtained. We have shown the character of the leaf which is required for chewing, and have found more clearly than ever that it is the quantity, and also the character, of the essential oil which seems most largely to determine the value of any sample of betel-vine leaf for this purpose. This being proved, the way is open for an attempt to discover what modification in the growing conditions will affect the amount and character of the essential oil in the desired direction—and this will be the next line of investigation as regards the cultivation of the vine.

The essential oil itself, however, is not always the same. It consists of a mixture of certain phenols and of certain terpene-like constituents. As far as the phenols are concerned, eugenol is always the chief constituent in Indian oils, mixed with a small quantity of betel phenol. We have never found chavicol in Indian oils. The best essential oil, from a point of view of public

taste, is that which contains as large a proportion of phenols as possible. Those varieties of leaf which gives an essential oil containing much terpene are very pungent, but are looked upon as very coarse. Bleaching not only increases the amount of essential oil in the leaf, but also increases the proportion of phenols in the essential oil. The nature of the terpene-like constituents is still unknown but wil be investigated at the first opportunity.

Bleaching of the leaf must be carried out in the dark, and can be most successfully accomplished at a higher temperature than is usual, say between 29° to 33°C. with much less loss, in much shorter time, and with a much more evenly bleached leaf than is usual. If over-bleaching is avoided, the increase in essential oil is nearly equal to that obtained in the ordinary process. The best means for carrying out this improved method in practice is now under investigation.

POONA,

October, 1915.

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